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Correlations Between Quantitative Indicators of Photosynthetic Pigments in *Vicia* Varieties under Conditions of Soil Salinization

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The article describes the effect of soil salinization on the level of correlations between quantitative indicators of photosynthetic pigments in vetch varieties. The objects of the study were the varieties of wiki Mirzachul-1 (*Vicia villosa* Roth) and Mirzachul-3 (*Vicia angustifolia*. L.). It was found that in highly saline soils, compared with weak ones, the amount of chlorophyll Chl «a» in the Mirzachul-1 variety decreased by 308 mg/g, the amount of chlorophyll Chl «b» and carotenoids (Car) increased by 0.11214 and 0.1300 mg/g.There was a strong positive correlation between Chl «a» and Chl «b» in all variants of the study. The similarity of correlation matrices is studied. At the same time, the similarity of correlation matrices between weakly and strongly saline soils was noted in the Mirzachul-1 variety by more than 90%, in the Mirzachul-3 variety by 69.1%. This indicates that the Mirzachul-1 varieties are stable in terms of the level of correlations according to the studied characteristics, which indicates a tendency to soil salinization.

Key words: Photosynthetic pigments. chlorophyll Chl «a», Chl «b», carotenoids, Car, salt resistance, correlation, levels of correlation bonds, soil salinity, similarity of correlation matrices

Photosynthetic pigments are responsible for absorbing, transmitting and converting light energy in photosynthesis. Quantitative and qualitative changes in pigments are a sensitive indicator of the physiological state of plants and the activity of the photosynthetic apparatus. In connection with global climate change and the exacerbation of regional environmental problems, increasing attention is being paid to the preservation of floristic wealth (Dymova & Golovko, 2007; Mokronosov, 2000).

The amount of pigments depends on the external environmental factor, primarily on light, as well as the length of daylight hours, air temperature, soil fertility, timing and sowing rate. Studies have shown that under extreme conditions Chl “a” is destroyed to a large extent. In the control, the content of Chl "a" in barley leaves was -0.590-0.720 mg/g after 46 hours of complete darkening -0.266-0.393 mg/g, and after 72 hours -0.181-0.339 mg/g. As a result, the chlorophyll content compared to the control decreased by 0.409-0.381 mg/g over 72 hours of darkening (Kakhnovich & Darwina 1997).

The quantity and quality of photosynthetic pigments depends on abiotic environmental factors. It was established that the presence of heavy metals and acetates in the cultivation medium: Ni^{2+}, Co^{2+}, Cu^{2+}, Pb^{2+} affected photosynthetic pigments, in particular chlorophylls Chl "a" and "b", which led to disruption of the photosynthesis process and a change in the ratio of chlorophylls Chl "a/b " According to the authors, exposure to heavy metals led to the degradation of Chl “a” in relation to chlorophyll “b” (Lobkova *et al.*, 2021).

The amount of photosynthetic pigments depends on the biological characteristics of plants, as well as cultivation conditions. Studying 31 species of steppe and forest plants in the Middle Urals, the authors came to the conclusion that steppe plants differed from forest plants in a lower content of chlorophylls and carotenoids in the leaf mass, as well as a low Chl/Car ratio (Ivanov *et al.*, 2020).

Studying the content of photosynthetic pigments of 160 plant species of the natural flora of the taiga zone of the European north-east of Russia, it was found that terrestrial plants of the taiga zone are characterized by a relatively low accumulation of chlorophylls, and the protective role of carotenoids increased with movement to the north. The amount of carotenoids in the leaves of terrestrial plants ranged from 0.2 to 5.1 mg/g, and was closely correlated with the chlorophyll content (r = 0.77), where the chlorophyll content ranged from 1 to 17 mg/g dry weight, herbaceous plants accumulated 1.5 times more chlorophyll, than woody ones (Dymova & Golovko, 2019).

Based on these data, the authors came to the conclusion that the study of photosynthetic pigments complements the characteristics of the floristic diversity of the climatic zone; under stressful conditions, the amount of carotenoids increases.

Agricultural practices, in particular, sowing rates, also influenced the amount of pigments. It was established that when the sowing rate of vetch cereal mixtures was increased in experiments to 5-6 and even 8 million germinating seeds per hectare, it inhibited the synthesis of pigments in hairy vetch leaves. Car resistance was superior to chlorophylls, while Chl “a” was superior to Chl “b” (Parakhin *et al.*, 2010). This means that with such an agrocenosis, unfavorable environmental conditions were created for the growth and development of vetch, as a result of which this led to a decrease in the amount of pigments.

The amount of chlorophyll per unit leaf area characterizes the adaptation of plants to the climatic conditions of a given geographical area, while the concentration of pigments per unit leaf mass demonstrates adaptation to local environmental conditions (Shirokikh *et al.*, 2022).

Based on these data, a brief conclusion can be drawn that the content of photosynthetic pigments can be considered as a sensitive factor of resistance to external, especially stressful environmental conditions. This is of great importance in conditions of soil salinity. High concentrations of salt (NaCl) in the soil cause stress in plants, which occurs in two stages. The first stage is the result of osmotic stress caused by a sharp drop in water potential in the root zone, and the second is caused by the toxic effects of the accumulation of salt ions directly in plant cells (Trofimov *et al.*, 2010).

One of the ways to rationally use saline soils is to use salt-tolerant crops. In this matter, hairy vetch are of great importance. According to literature data, the protein content in dry mass is 20-25%, in grain – over 25%. 100 kg of green mass contains 16 feed units and more than 2.5 kg of digestible protein. Along with its nutritional benefits, hairy vetch plays an important agrotechnical role; it helps clear crops of weeds and leaves up to 100 kg/ha of biological nitrogen. The proportion of Chl “a” content in hairy vetch leaves is 73-78%, Chl “b” 27-22% (Parakhin *et al.*, 2010).

Hairy vetch is recognized as a salt-tolerant crop (Trofimov *et al.*, 2010). It should be noted that in the conditions of the Republic there are 10 types of vetch. Among these species, the most interesting are hairy vetch (*Vicia villosa* Rot), common vetch (*Vicia sativa* L), and narrow-leaved vetch (*Vicia angustifolia.* L). Studying vetch under conditions of soil salinity, it was found that hairy vetch as a forage, intermediate crop is of great interest and is an important component of the agrocenosis (Kuliev *et al.*, 2020; Ismoilova *et al.*, 2023).

The correlation between photosynthetic pigments and economic traits was found to be positive between the level of winter hardiness of vetch and the content of tightly bound forms of pigments. The correlation coefficient between the amount of tightly bound pigments in plants in winter and their frost resistance was 0.583, and winter hardiness was 0.678 (Parakhin *et al.*, 2010; Telichko & Mokhan, 2018).

According to the literature data, it can be said that the amount of photosynthetic pigments complements the characteristics of the floristic diversity of the climatic zone. With their help, you can predict productivity and resistance to environmental conditions. Especially the content of carotenoids provides great information about the resistance of genotypes. But the amount of photosynthetic pigments under soil salinity conditions and the correlations between pigments have not been studied. In this regard, we were given the task of studying the influence of the level of soil salinity on the correlations of photosynthetic pigments in vetch varieties.

MATERIALS AND METHODS

The experiment was carried out in saline (weak, medium and strong) soil conditions at the experimental site of Gulistan State University. The objects of the study were hairy vetch (*Vicia villosa* Roth) and narrow-leaved vetch (*Vicia angustifolia*. L). Method for quantitative determination of chlorophyll: about 1.0 g (exactly weighed) of crushed raw material is placed in a 100 ml heat-resistant flask with a reflux condenser and extracted with ethyl alcohol 96% (30 ml each) in a boiling water bath for 30 minutes. The extraction is repeated two more times. The resulting extracts are filtered into a 100 ml volumetric flask and adjusted to the mark with 96% ethyl alcohol. After cooling, 5 ml of the resulting extract is transferred to a 25 ml volumetric flask and adjusted to the mark with 96% ethyl alcohol. The reference solution is ethyl alcohol 96%. The content of Chl "a", Chl "b" and carotenoids (Car) in the leaves was measured by optical density using an SF-2000 spectrophotometer at wavelengths of 662 and 644 nm, respectively, for chlorophylls "a" and "b", 470 nm - for carotenoids (Maslova *et al.*, 1986). The content of pigments in leaves was expressed in mg/g dry weight (Strusovskaya, 2012; Maslova *et al.*, 1986).

Statistical data processing was carried out using the SPSS-17 program. With the help of this program, static indicators of the studied characteristics, the level of correlations were calculated, and factor analysis was carried out. To assess the average level of connections, the coefficient of determination (the square of the correlation coefficient – r^2) is used. The indicator of similarity of matrices by structure was used by the correlation coefficient between z-transformed matrices (r_z; R. Fisher's z-transformation is introduced to bring the distribution closer to normal). To compare the correlation matrices, the following gradations were used: if the similarity of the correlation matrices is 90% or more, they are similar; below 80-75% - the differences are significant (Rostova, 2002; Kuliyev *et al.*, 2021)

RESULTS AND DISCUSSION

The studied vetch varieties Mirzachul-1 and Mirzachul-3 differed in the content of photosynthetic pigments. Chlorophyll Chl “a” on average in the

Mirzachul-1 variety was 3.00 mg/g, Chl “b” - 1.62 mg/g, the ratio of these pigments was 1.80, the total content of these pigments was 4.62. The amount of carotenoids (Car) in the Mirzachul-1 variety was 0.68, and in the Mirzachul-3 variety it was 2.12 mg/g. In conditions of slightly saline soil, the Mirzachul-3 variety was superior in carotenoid content and lower in chlorophyll content. On moderately saline soils, most of the pigments studied in the Mirzachul-1 variety were superior to Mirzachul-3. Chlorophyll Chl “a” in the Mirzachul-1 variety was 2.94, and in the Mirzachul-3 variety it was 1.28 mg/g. Only in terms of Car content, the Mirzachul-3 variety was superior to Mirzachul-1. On slightly and moderately saline soils, the content of carotenoids was higher in the Mirzachul-3 variety, and on highly saline soils it was inferior to the Mirzachul-1 variety.

All parameters of photosynthetic pigments decreased in highly saline soils (Fig. 1). Compared to slightly saline soils, the amount of Chl “a” decreased by 0.308 mg/g or 10.27%, Chl “b” increased by 0.1214 mg/g (7.48%). The content of carotenoids also increased under conditions of highly saline soils by 0.1300 mg/g (18.99%).

The maximum amount of pigments was contained in the leaves. In the Mirzachul-1 variety on slightly saline soils, the content of Chl “a” was maximum – 5.25 mg/g (77.35%) in leaves, 0.95 mg/g (13.94%) in stems, 0.57 mg/g (8.40%) in flowers and 0.2 (0.29%) beans. Similar results were obtained on moderately saline and highly saline soils. Our data is consistent with other data. It has been established that hairy vetch leaves contain 73-78% chlorophyll “a” and 27-22% chlorophyll “b” (Parakhin *et al.*, 2010).

Correlation analyzes provide information about the relationship between the studied characteristics. Study indicators of photosynthetic pigments correlate with morphological characteristics of plants and abiotic environmental factors. It was established that the relationship between the frost resistance of vetch and the quantitative content of tightly bound chlorophyll and carotenoids in winter is positive, where the correlation coefficient was r = 0.428 and 0.520. Positive correlations have been established between winter hardiness and the number of tightly bound forms of pigments (r = 0.577 and 0.533) (Parakhin *et al.*, 2010).

In conditions of slightly saline soil, a high correlation was noted between the amount of Chl "a" (1) and chlorophyll Chl "b" (2), as well as the Chl/Car ratio (5), where the correlation coefficient is r => 0.7 (Fig. 2). This means that the more Chl “a” and the more Chl “b”. An increase in the amount of chlorophylls leads to an increase in their proportional ratio. There is also a strong correlation between the characters Chl “a/b” (3) and Car (4). An increase in the amount of carotenoids led to an increase in chlorophylls. A weak negative correlation was noted between the amount of Car (4) and the proportion of Chl/Car(5). In such cases, an increase in the amount of carotenoids leads to a decrease in the Chl/Car ratio. It should be noted that positive correlations were noted between the amount of Chl “a” (1) and Chl “b” (2), as well as the ratio of Chl on slightly and highly saline soils. An increase in the amount of chlorophylls led to an increase in the proportion of Chl/Car. In addition, a weak negative correlation was established between the content of carotenoids (4) and the Chl/Car ratio (5) in all variants of the study. This indicates that as Car increases, the fractional ratio Chl/Car (5) decreases.

In the vetch variety Mirzachul-3, a strong correlation was established between chlorophyll (Chl) “a” (1) and Chl “b” (2) and the amount of Car(4) in all variants of the study (Fig. 3). We remind you that in the Mirzachul-1 variety, weak correlations were established between chlorophylls Chl) “a” (1) and Chl “b” (2) with carotenoids Car (4). This means that in the Mirzachul-3 variety these connections were strengthened. On weakly and moderately saline soils, a strong positive correlation was established between characteristics 3 and 5; with strong salinity, a strong but negative correlation was established between these indicators. This means that an increase in the Chl “a/b” ratio leads to an increase in the Chl/Car ratio. This is due to a decrease in the amount of carotenoids in the Mirzachul-3 variety in highly saline soils (Table 1). According to the tabular data, it is clear that in the Mirzachul-3 variety the amount of carotenoids on slightly and moderately saline soils was 2.12-1.14 mg/g, and on highly saline soils - 0.75 mg/g. This means that an increase in the level of soil salinity led to a decrease in the amount of carotenoids in the Mirzachul-3 variety.

Based on these data, we can say that soil salinity influenced the level of correlations. Especially, strong changes occurred in the Mirzachulm -3 variety (Fig. 3).

To study the influence of the level of soil salinity on the structure of correlations, we used a new method that was developed by N.S. Rostova (2002). The essence is that if the similarity of correlation matrices is more than 90%, they are similar, less than 90% indicates a change in the structure of connections. According to our data, the Mirzachul-1 variety in all variants of the study had a similarity coefficient of more than 90%, which indicates the similarity of these matrices. In such cases, the Mirzachul-1 variety is relatively prone to the level of soil salinity compared to the Mirzachulm-3 variety, where the coefficient of similarity between slightly saline and highly saline soils was 69.1%, which indicates a strong difference between the matrices (Table 2). This evidences that the level of soil salinity significantly affected the degree of correlations in the Mirzachul-3 variety. As a result, in conditions of highly saline soils, changes occurred in the level of correlations.

Table 1: Effect of soil salinity level on the amount of photosynthetic pigments in vetch varieties

	Chl «a»	**Chl** «b»	**Chl** «a/b»	**Chl «a»+** «b»	**Car**	***Chl*/Car**
	In conditions of slightly saline soils					
Mirzachul -1	3,00±0,84	1.62±0.46	1,80±0.07	4.62±1,30	0.68±0.19	12,20±4,48
Mirzachul- 3	2,35±0,8	2,31±0.80	1,13±0.06	4.66±1,61	2,12±0.73	2,49±0,18
	In conditions of moderately saline soils					
Mirzachul -1	2.94±0.83	1,65±0.34	1,64±0,10	4,59±1,29	0,77±0.22	9,83±3,25
Mirzachul- 3	1,28±0.44	1,33±0.45	0,92±0,02	2,61±0,89	1,40±0.47	1,82±0,03
	In highly saline soil conditions					
Mirzachul -1	2.69±0.75	1.74±0.48	1,41±0.10	4.43±1,24	0,81±0.23	8,68±2,81
Mirzachul- 3	0,64±0,18	0,77±0.20	0,84±0.06	1,71±0,38	0,75±0.21	2,31±0,55

Note: chlorophyll “a”- Chl “a”; Chlorophyll “b”-Chl “b”,Chl “a/b” chlorophyll ratio. Car-carotenoids; Chl/Car ratio,

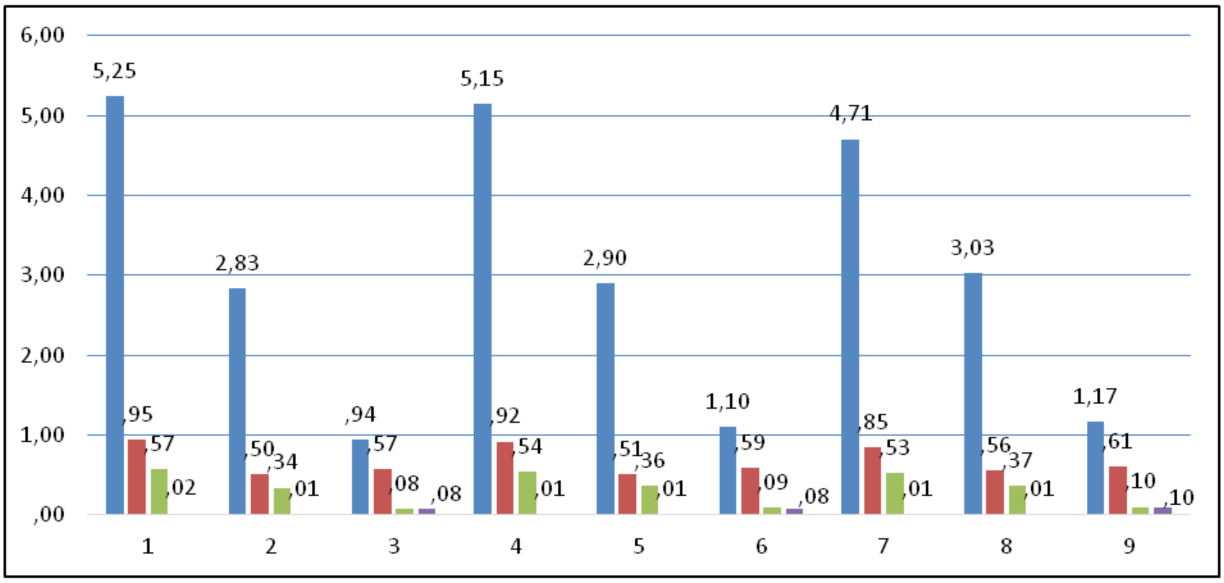

Figure 1: Content of photosynthetic pigments on the morphological organs of vetch (Variety Mirzachulm-1): Note: the first column is the content of chlorophyll “a” in the leaves; the second is the stem; the third is a flower; the fourth is a bean. The numbers indicate the level of soil salinity: 1, 2 and 3 — slightly saline; 4, 5 and 6 - moderately saline; 7, 8 and 9 highly saline soils

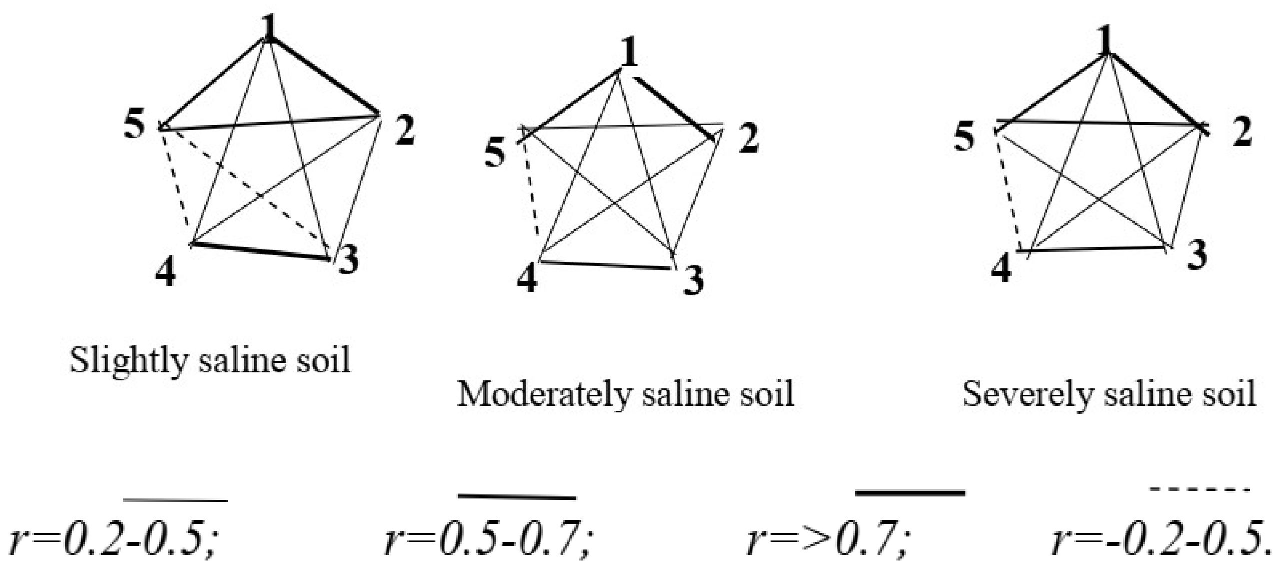

Figure 2: The influence of soil salinity on the degree of correlations in the vetch variety Mirzachul-1

Note: the numbers indicate the characteristics of 1 - chlorophyll-(Chl) “a”; 2 - chlorophyll-(Chl) "b"; 3 - chlorophyll (Chl) ratio “a”/“b”; 4 - carotenoids; 5 - ratio Chl/Car carotenoids.

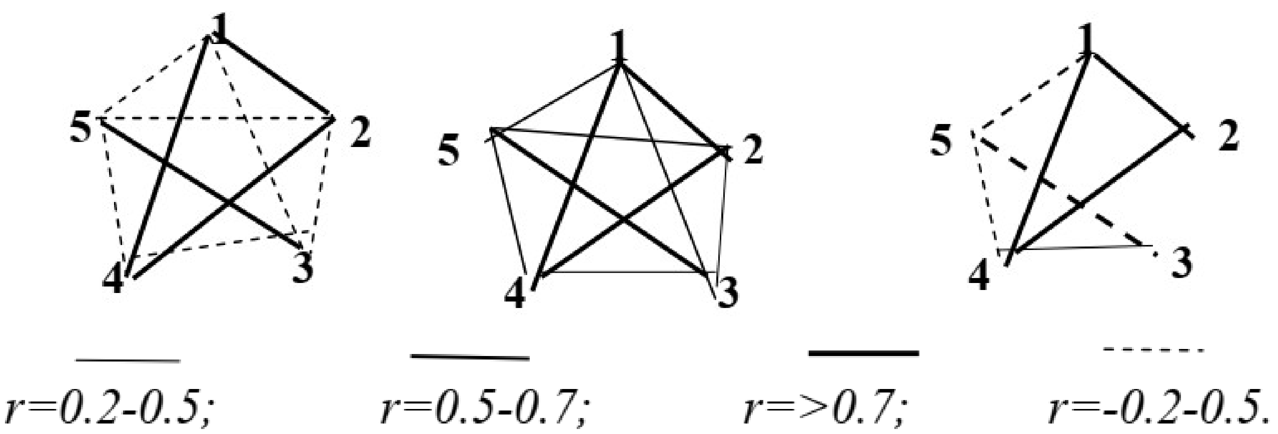

Figure 3: The influence of soil salinity on the degree of correlations in the vetch variety Mirzachul-3

Note: the numbers indicate the characteristics of 1 - chlorophyll-(Chl) “a”; 2 - chlorophyll-(Chl) "b"; 3 - chlorophyll (Chl) ratio “a”/“b”; 4 - carotenoids; 5 - ratio Chl/Car carotenoids.

Table 2: Similarity of correlation matrices among vetch varieties

vetch varieties	**Soil salinity level**	**vetch variety Mirzachul-1**			**vetch variety Mirzachul-3**		
		Soil salinity level					
		Slightly	***Moderately***	***Severely***	***Slightly***	***Moderately***	***Severely***
		Similarity of correlation matrices, %					
Mirzachulm-1	*Slightly*	-	98,3	99,2	47,6	40,4	38,2
	Moderately			99,0	44,9	36,1	31,9
	Severely				45,4	36,4	31,5
Mirzachul-3	*Slightly*					96,1	69,1
	Moderately						81,5
	Severely						-

CONCLUSION

The level of soil salinity affected the amount of photosynthetic pigments. An increase in the level of soil salinity led to a decrease in the amount of Chl “a” in the Mirzachul-1 variety by 0.308 mg/g or 10.3%, Chl “b” increased by 0.1214 and Car by 0.1300 mg/g compared to slightly saline soils . In the Mirzachul-3 variety, in contrast to the Mirzachul-1 variety, Chl “a” decreased by 72.77% on slightly saline soils compared to 10.3% in the Mirzachul-1 variety. In addition, Chl “b” decreased by 66.57% and Car by 64.42%. According to these data, the Mirzachul-3 variety was severely damaged by the action of soil salts. This gives us reason that the Mirzachul-1 variety is the most resistant to soil salinity compared to the Mirzachul-3 variety. According to literature data, it is known that under stressful conditions the amount of carotenoids increases. This is one of the mechanisms of plant adaptation to stressful conditions. Under conditions of soil salinity, a similar phenomenon occurs. This means that the vetch variety Mirzachul-1, which belongs to the hairy vetch species, can be said to be tolerant to salt conditions, similar conclusions were made by other researchers (Kuliev *et al.*, 2020).

Thus, based on the results obtained, we can draw the following conclusions.

1. Under soil salinity conditions, a strong (r=>0.70) correlation was established between the chlorophylls Chl “a” and Chl “b” in both vetch varieties, as well as in all research variants.

2. When the level of soil salinity increased, the amount of chlorophyll Chl “a” decreased. The amount of chlorophyll Chl "b" and carotenoids depended on the level of soil salinity, as well as on the genotype.

3. Vetch varieties Mirzachul-1 which belong to hairy vetch (Vicia villosa Roth) are recognized as salt-tolerant varieties.

CONFLICTS OF INTEREST

The authors declare that they have no potential conflicts of interest.

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